

neuroepithelium during eye morphogenesis and in retinal progenitor cells and photoreceptors during retinogenesis. We have isolated a 2.8 kb fragment of regulatory DNA from the *Rx2A* gene. We have identified a forkhead binding site in this genomic region. Our objective is to determine the role that eye-specific forkhead transcription factors play in *Rx* transcriptional regulation during eye development. We introduced *Rx2A*/GFP transgenes into *X. laevis* embryos via intracytosolic sperm injections. The full-length *Rx2A*/GFP transgene recapitulates the expression pattern of the endogenous *Rx2A* gene. We identified eye-specific forkhead factors and confirmed their expression in *X. laevis* by in situ hybridization using specific antisense riboprobes. We tested candidate forkhead factors for binding specificity using electrophoretic mobility shift assays. We have demonstrated that a putative forkhead binding site is essential for normal promoter activity using transgenic embryos. We found that the expression patterns of *FoxD1*, *FoxN4*, *FoxO3* and *FoxM1* overlap *Rx* expression in the developing eyes of *X. laevis* embryos. We demonstrate that *FoxN4* and *FoxO3* bind to the putative forkhead site within the *Rx2A* promoter. Our data implicate retinal forkhead transcription factors as regulators of *Rx* transcription during eye development.

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Decoding *cis*-regulatory sequences involved in coordinate gene expression in *Drosophila*

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Understanding the regulatory mechanisms that control coordinate gene expression is a longstanding goal of biology. We have recently reported the development of EvoPrinter (Proc. Natl. Acad. Sci. 102; 14700–5, 2005: <http://evoprinter.ninds.nih.gov/>), a phylogenetic footprinting tool that identifies multi-species conserved sequences (MCSs) within a reference DNA. Within known *cis*-regulatory regions, many of the MCSs contain motifs that are identified binding sites of known transcriptional regulators. Additional parsing functions have been added to the basic EvoPrinter algorithm to facilitate the breakdown of the MCSs into shorter sequences that allow for sequence comparison between enhancers. *cis*Decoder is a second algorithm we have developed to analyze and compare enhancers. EvoPrinter and *cis*Decoder have been applied to identified enhancers of *Drosophila* neural precursor genes and mesodermal determinants. Comparison of these two allow us to distinguish between enhancer elements that are common and those that are specific to enhancer specificity. In

this comparison, shared sequence elements were found to be overlapping or adjacent to known transcription factor DNA-binding sites. The function of many of these elements shared by enhancers of co-expressed genes is as yet unknown. The results thus far suggest that we can begin to decode key *cis*-regulatory sequences involved in coordinate gene regulation.

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The level of CaM kinase II is higher in brains of learning-enhanced *Drosophila melanogaster*

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The multifunctional calcium/calmodulin-dependent protein kinase II (CaM kinase II) is a major protein that coordinates cellular responses to external cues such as hormones and nerve signals. In rodent and human hippocampus, the primary role of CaM kinase II is to participate in memory formation such as spatial learning. CaM kinase II has been isolated from the brains of *Drosophila melanogaster* and is known to function in nerve signal transduction in the fly. However, how CaM kinase II carries out its action has not been extensively studied. More recently, it has been shown that memory formation in the fly is light-dependent. We cultured wild-type *D. melanogaster* under different lighting conditions that influenced their learning. Using immunoblotting and chemiluminescence detection, we found that the brains of the flies grown under conditions that facilitated learning expressed higher levels of CaM kinase II.

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Transcriptional and post-transcriptional regulation of the *nerfin-1* expression during *Drosophila* neurogenesis

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nerfin-1 belongs to a conserved subfamily of Zn-finger transcription factors. *nerfin-1* is required for interneuron axon guidance (Kuzin et al., Dev. Biol. 277: 347–65, 2005). During embryonic CNS development, *nerfin-1* mRNA is detected in all early delaminating neuroblasts, many GMCs and transiently in most, if not all, nascent neurons. However, the nuclear Nerfin-1 protein is detected only in neural precursor cells that undergo a single final division to generate neurons (the MP neuroblasts and GMCs) and is